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Acute Toxicity of Ten Chlorinated Aliphatic Hydrocarbons to the Fathead Minnow (*Pimephales promelas*)

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Abstract. Ninety-six-hour LC50 values were determined for 10 chlorinated aliphatic hydrocarbons in freshwater flow-through toxicity tests on fathead minnows (*Pimephales promelas*). The 96-hr measured LC50 values in mg/L from combined duplicate tanks were: tetrachloroethylene 13.4; 1,1′,2,2′-tetrachloroethane 20.4; pentachloroethane 7.34; 1,1′,2-trichloroethane 81.6; hexachlorobutadiene 0.10; 1,1′2-trichloroethylene 45.0; 1,2-dichloropropane 140; 1,2-dichloroethane 116; hexachloroethane 1.51; 1,3-dichloropropane 131.

The compounds were chosen for study; [1] as part of the development of water quality criteria listed under section 307 (a) (1) of the U.S. Clean Water Act of 1977 (P.L. 95–217); [2] in partial satisfaction of paragraph 11 of the settlement agreement in *Natural Resources Defense Council et al. vs Train,* 8 Environmental Reporter Case (District of the District of Columbia 1976); and [3] as part of the ongoing effort at the Environmental Research Laboratory-Duluth to relate chemical structure to biological activity. A structure-activity data base is being developed from the biological and physical activity of tested compounds. This data base is intended for use in making predictions of toxic and environmental effects of untested compounds.

The chlorinated ethanes tested were 1,1′,2,2′-tetrachloroethane, pentachloroethane, 1,1′,2-trichloroethane, 1,2-dichloroethane, and hexachloroethane. These compounds are used for metal cleaning in industry, as solvents for waxes, oils, and resins, and as chemical feedstocks (Hawley 1977).

Perchloroethylene (1,1′,2,2′-tetrachloroethylene) is used primarily as a solvent in the dry cleaning industries, and to a lesser extent as a degreaser in metal industries, and it reaches receiving waters in various effluents. However, its level there is minimal because of its high volatility. Nonetheless, it has been reported in trace amounts in U.S. and English waters and in aquatic organisms, air, and foodstuffs in England. Persistence in the environment is negligible because of photo-oxidation and attack by hydroxyl and chlorine free radicals (U.S. Environmental Protection Agency 1980d).

Hexachlorobutadiene is used as a solvent for elastomers, as a heat transfer liquid, as a transformer and hydraulic fluid, and as a wash liquor for removing hydrocarbons. Its release into the environment is primarily through wastes from chlorinated-hydrocarbon-using industries. While it volatilizes readily from water, it has been reported in drinking water supplies in the U.S. Sorption may be important with this compound, since it tends to concentrate in sediments (U.S. Environmental Protection Agency 1980c).

Trichloroethylene is used as a degreasing solvent in metal industries. Other uses are in household and industrial dry cleaning solvents and as an extractive solvent in food processing. Volatilization losses of this chemical are the major sources of environmental contamination. This compound has been found in air, food, human tissues, rivers, municipal water supplies, the sea, and in aquatic organisms. However, it is not expected to persist in the environment because of its rapid photo-oxidation, high volatility, and low water solubility (U.S. Environmental Protection Agency 1980e).

The uses of 1,2-dichloropropane and 1,3-dichloropropane are as soil fumigants for nematode control, as oil and fat solvents, and as dry cleaning agents. These compounds reach the environment

Table 1. Aquatic toxicities of tested compounds

Compound	Organism	Effect measured	Value	Source
Tetrachloro-ethylene	*Pimephales promelas* Fathead minnow	embryo larval MATC[a]	1.4 – 2.8 mg/L	DeFoe (unpublished data)
	Daphnia magna Water flea	48 hr LC50	9.09 – 18.1 mg/L	
		EC50[b]	7.49 – 8.50 mg/L	
		NOEC[c] for both growth and reproduction	.505 mg/L	Richter (unpublished data)
		24 hr LC50	18 mg/L	LeBlanc (1980)
		48 hr LC50	18 mg/L	
		no discernible effect concentration	10 mg/L	
	Lepomis macrochirus Bluegill	21 day BCF[d] in water concentration of 3.43 mg/L	49 mg/L	Barrows *et al.* (1980)
1,1',2,2'-Tetra-chloroethane	*P. promelas*	embryo-larval MATC	1.4 – 4.0 mg/L	DeFoe (unpublished data)
	D. magna	48 hr LC50	56.9 – 62.1 mg/L	Richter (unpublished data)
		EC50	23.0 – 25.2 mg/L	
		NOEC (for reproduction)	6.85 mg/L	
		24 hr LC50	18 mg/L	LeBlanc (1980)
		48 hr LC50	9.3 mg/L	
		no discernible effect concentration	<1.7 mg/L	
	L. macrochirus	14 day BCF in water concentration of 9.62 μg/L	8	Barrows *et al.* (1980)
Pentachloro-ethane	*P. promelas*	embryo-larval MATC	0.9 – 1.4 mg/L	DeFoe (unpublished data)
	D. magna	48 hr LC50	7.32 – 8.02 mg/L	Richter (unpublished data)
		EC50	4.69 – 6.88 mg/L	
		24 hr LC50	63 mg/L	Leblanc (1980)
		48 hr LC50	63 mg/L	
		no discernible effect concentration	43 mg/L	
	L. macrochirus	14 day BCF in water concentration of 7.93 μg/L	67	Barrows *et al.* (1980)
1,1',2-Tri-chloroethane	*P. promelas*	embryo-larval MATC	6.0 – 14.8 mg/L	DeFoe (unpublished data)
	D. magna	48 hr LC50	174 –186 mg/L	Richter (unpublished data)
		EC50	77.8 – 80.6 mg/L	
		chronic no observable effect concentration	26.0 mg/L	
		reproduction, growth	13.2 mg/L	
		24 hr LC50	19.0 mg/L	LeBlanc (1980)
		48 hr LC50	18.0 mg/L	
		no discernible effect concentration	1.0 mg/L	
		48 hr LC50	43 mg/L	Adema (1978)
	Orphyryotrocha labronica Marine polychaete	Shock 96 hr LC50 (transfer with no acclimation to intermediate levels of toxicant)	160 mg/L	Rosenberg *et al.* (1975)
Hexachloro-butadiene	*P. promelas*	embryo-larval MATC	.0065– .013 mg/L	Benoit *et al.* (1982)
	Poecilia latipinna Sailfin molly	96 hr LC50	1.6 mg/L	Laska *et al.* (1978)
	Brachydanio rerio Zebrafish	48 hr LC50	1 mg/L	Sloof (1979)
	Salmo gairdneri Rainbow trout	detection limit of biological monitoring system	.05 mg/L	Sloof (1979)

Table 1. (cont'd)

Compound	Organism	Effect measured	Value	Source
1,1',2-Trichloroethylene	*P. promelas*	96 hr LC50	40.7 mg/L	Alexander *et al.* (1978)
	D. magna	48 hr LC50	42 – 97 mg/L	Canton and Adema (1978)
	D. pulex	48 hr LC50	45 mg/L	
	D. cucullata	48 hr LC50	57 mg/L	
	D. magna	24 hr LC50	22 mg/L	LeBlanc (1980)
		48 hr LC50	18 mg/L	
		no discernible effect concentration	2.2 mg/L	
	S. gairdneri	detection limit in biological monitoring system	5 mg/L	Sloof (1979)
1,2'-Dichloropropane	*P. promelas*	embryo-larval MATC	6 – 11 mg/L	Benoit *et al.* (1982)
	D. magna	24 hr LC50	99 mg/L	LeBlanc (1980)
		48 hr LC50	52 mg/L	
		no discernible effect concentration	22 mg/L	
	L. macrochirus	96 hr LC50	320 mg/L	Dawson *et al.* (1975/77)
1,2-Dichloroethane	*P. promelas*	embryo-larval MATC	29 – 59 mg/L	Benoit *et al.* (1982)
	D. magna	48 hr LC50	268 –315 mg/L	Richter (unpublished data)
		EC50	155 –183 mg/L	
		chronic no observable effect concentration		
		reproduction	10.6 mg/L	
		growth	41.6 mg/L	
	D. magna	24 hr LC50	250 mg/L	LeBlanc (1980)
		48 hr LC50	220 mg/L	
		no discernible effect concentration	<68 mg/L	
	O. labronica	Shock 96 hr LC50	400 mg/L	Rosenberg *et al.* (1975)
		Acclimated 96 hr LC50	900 mg/L	
	L. macrochirus	96 hr LC50	550 mg/L	Dawson *et al.* (1975-77)
	L. macrochirus	14 day bioconcentration factor at 95.6 μg/L	2 mg/L	Barrows *et al.* (1980)
	Gammarus fasciatus amphipod	96 hr LC50	>100 mg/L	Johnson and Finley (1980)
	Pteronarcys sp. stonefly	96 hr LC50	>100 mg/L	
	S. gairdneri	96 hr LC50	225 mg/L	
Hexachloroethane	*P. promelas*	embryo-larval MATC	0.069 – 0.207 mg/L	DeFoe (unpublished data)
	D. magna	48 hr LC50	2.90 – 2.35 mg/L	Richter (unpublished data)
		EC50	1.81 – 2.10 mg/L	
	D. magna	24 hr LC50	26 mg/L	LeBlanc (1980)
		48 hr LC50	8.1 mg/L	
		no discernible effect concentration	0.28 mg/L	
	L. macrochirus	BCF 28 days at water concentration of 6.17 μg/L	139 mg/L	Barrows et al. (1980)
1,3-Dichloropropane	*P. promelas*	embryo-larval MATC	8 – 16 mg/L	Benoit *et al.* (1982)
	D. magna	24 hr LC50	490 mg/L	LeBlanc (1980)
		48 hr LC50	280 mg/L	
		no discernible effect concentration	68 mg/L	

[a] Maximum acceptable toxicant concentration
[b] Effect ruled was inability to swim; animals were alive but incapacitated
[c] No observable effect concentration
[d] Bioconcentration factor

via agricultural run-off as well as via industrial and municipal effluents.

A review of the literature dealing with aquatic toxicities of the tested compounds is summarized in Table 1. More extended background data for these compounds is available in the Criteria Documents (U.S. Environmental Protection Agency 1980a–1980e).

Materials and Methods

Exposure System

Two 2-L/cycle proportional diluters with dilution factors of 0.6 (Mount and Brungs 1967) were used to deliver five toxicant concentrations and a control, in duplicate, to randomly arranged exposure chambers. These chambers were all-glass aquaria with a working volume of 41 L, 30 cm high by 30 cm by 60 cm, with a standpipe topped with a cylinder of stainless-steel screen to prevent loss of fish. Flows were greater than ten tank-volumes per day. Fluorescent lighting on a 16 hr photoperiod was used, and the light level was 48 lumens at the water surface.

Physical and Chemical Conditions

The laboratory lake water supply (Lake Superior) was the source of the dilution water. This was heated to maintain a mean of 25°C with a standard deviation of 0.7°C in the test tanks. At least once during each 4-day test, pH, dissolved oxygen (DO), hardness (as $CaCO_3$), and alkalinity (as $CaCO_3$) were determined for control, an intermediate, and the high tanks. The pH ranged from 6.7 to 7.6. Means and ranges for DO, hardness, and alkalinity, were 8.0 mg/L (7.6–9.2), 45.1 mg/L as $CaCO_3$ (45.0–45.5), and 41.8 mg/L as $CaCO_3$ (35.6–43.4), respectively, for all tests. Values obtained from high exposure concentrations did not deviate significantly from those observed in the controls. Water chemistry methods were those recommended by the American Public Health Association (APHA *et al.* 1980) and the U.S. Environmental Protection Agency (USEPA 1974).

Biological Methods

Laboratory-reared fathead minnows (*Pimephales promelas*), 30 to 35 days old, were used in these experiments. The rearing water was the same as the diluent water, with the temperature held at a temperature of 25° ± 2°C. Fish in the rearing tanks were fed live brine shrimp nauplii in excess until 12 to 24 hr before testing, then not fed during the exposure period.

At the beginning of a flow-through test, 50 fish were randomly assigned to each of the 12 exposure tanks. Dead fish were counted and removed at least twice during the first day, and twice daily after that. A saturator system was used to solubilize the chemicals (Phipps *et al.* 1982). This saturator uses an intermittent flow of water, delivered by a metering pump, through one or more closed chambers (19 L stainless steel soda carbonation tanks). The water is continuously mixed with the toxicant, the latter was added in a single charge before the test began.

Methods not discussed here followed those specified by the U.S. Environmental Protection Agency (1975).

Table 2. Gas chromatograph column temperatures and retention times

Compound	Temperature (°C)	Retention time (min.)
1,1′,2-Trichloroethane	40	1.63
1,1′,2,2′-Tetrachloroethane	40	1.65
Tetrachloroethylene	60	1.55
Pentachloroethane	40	1.92
Hexachloroethane	80	2.21
Hexachlorobutadiene	80	4.90
1,2-Dichloroethane	40	3.93
1,2-Dichloropropane	40	3.80
1,3-Dichloropropane	40	2.29
1,1′,2-Trichloroethylene	40	3.12

Chemical Methods

Chemical analyses for 1,1′,2-trichloroethane, 1,1′,2,2′-tetrachloroethane, tetrachloroethylene, pentachloroethane, hexachloroethane, and hexachlorobutadiene were performed with a Hewlett-Packard 5730A gas chromatograph equipped with a Model 3552A data system and a ^{63}Ni electron capture detector. The column was packed with 100/120 mesh Supelcoport® coated with 1.5% SP2250/1.95% SP-2401. A Tracor MT-220 manual gas chromatograph with a ^{63}Ni electron capture detector was used for analyzing 1,2-dichloroethane, 1,2-dichloropropane, 1,3-dichloropropane, and 1,1′,2-trichloroethylene. The column was packed with 80/100 mesh Gas-Chrom Q® coated with 4% SE-30/6% OV-210. The carrier gas for all compounds was 5% methane in argon, and the column temperatures and retention times are given in Table 2.

Fifty ml hexane was added to 100 ml volumetric flasks, which were then brought up to the mark with the water sample. Samples were stirred vigorously on magnetic stirrers for 1.5 hr and the organic layer was allowed to separate for 1 hr. Samples were diluted with hexane if necessary and analyzed as outlined above by comparison to known hexane standards. Known amounts of the chemicals were added to water samples to determine the efficiency of this extraction procedure. The over-all recovery for all compounds was 99 ± 4% (n = 69). Final results were not corrected for recovery.

Statistical Methods

The LC50 concentrations were calculated by using the Trimmed Spearman-Karber method for estimating median lethal concentrations (Hamilton *et al.* 1977). This method is not subject to the deficiencies of the more common normal or logistic methods and it is not as sensitive to anomalous responses on the conventional Spearman-Karber technique.

Results

The 24-, 48-, 72-, and 96-hr LC50 values and 95% confidence intervals of the chlorinated aliphatic compounds are given in Table 3. The 96-hr LC50 values ranged from 0.10 mg/L for hexachlorobuta-

Table 3. LC50 values (mg/L) of the chlorinated compounds

Compound	24 hr LC50	48 hr LC50	72 hr LC50	96 hr LC50
Tetrachloroethylene	17.9 (17.3–18.4)[a]	15.9 (15.0–16.8)	14.9 (13.9–15.8)	13.4 (12.4–14.4)
1,1′,2,2′-Tetrachloroethane	22.8 (21.9–23.8)	22.2 (21.2–23.1)	20.4 (20.0–20.8)	20.4 (20.0–20.9)
Pentachloroethane	7.72 (7.45–7.99)	7.43 (7.16–7.71)	7.34 (7.07–7.63)	7.34 (7.07–7.63)
1,1′,2-Trichloroethane	81.6 (60.9–109)[b]	81.6 (60.9–109)[b]	81.6 (60.9–109)[b]	81.6 (60.9–109)[b]
Hexachlorobutadiene	—	0.23 (0.20–0.26)	0.13 (0.09–0.18)	0.10 (0.09–0.11)
1,1′,2-Trichloroethylene	58.8 (57.8–59.7)	57.9 (57.2–58.6)	55.4 (53.0–57.8)	45.0 (41.9–48.4)
1,2-Dichloropropane	194 (184–205)	154 (144–166)	141 (132–151)	140 (131–150)
1,2-Dichloroethane	141 (131–153)	118 (111–125)	116 (110–123)	116 (110–123)
Hexachloroethane	1.80 (1.70–1.91)	1.55 (1.47–1.63)	1.55 (1.47–1.63)	1.51 (1.43–1.58)
1,3-Dichloropropane	133 (126–139)	131 (124–137)	131 (124–137)	131 (124–137)

[a] 95% confidence limits

[b] It was not possible to calculate confidence limits with the Spearman-Karber method. The intervals shown are derived from the binomial method and represent greater than 95% confidence level

diene to 140 mg/L for 1,2-dichloropropane. With the exception of hexachlorobutadiene, the effect of these compounds is narcotic; lethargy and anesthesia are observed. The other directly observable effect of hexachlorobutadiene is hemorrhaging. With the nine strictly narcotic-effect compounds, the toxicity increases with increasing chlorination and double bonding. Since a double bond deletes two hydrogens the toxicity goes up directly with the ratio of chlorine to total molecular weight.

When all the 96-hr LC50s are compared in terms of molar concentration, the relative toxicities range from 4×10^{-7} moles for hexachlorobutadiene to 1×10^{-3} moles for 1,2-dichloropropane.

The 1,1′,2-trichloroethane results were unusual in that all of the deaths that were to occur during the four days of the test happened in the first five hours.

The hexachlorobutadiene 24-hr LC50 is not available because in the highest concentration tested, 0.28 mg/L, 50% of the animals had not died after 24 hours.

Discussion

The data on these chemicals, as published in the toxicity literature, are difficult to use for comparison. For example, how is a 96-hr LC50 related to the 14 day bioconcentration factor (BCF)? However, invertebrates appeared more resistant to these chemicals than fish, with the exception of 1,1′,2-trichloroethylene where toxicities are about the same in fathead minnows and daphnids. The results with 1,2-dichloropropane indicated the fathead is substantially more resistant than *Daphnia magna,* as reported by LeBlanc (1980).

Data for different species of fish were, in some cases, comparable. Hexachlorobutadiene had a fathead minnow 48-hr LC50 of 0.23 mg/L in the present study compared with 1 mg/L for the zebrafish (Sloof 1979). The 96-hr LC50 for hexachlorobutadiene was 0.10 mg/L compared with 1.6 mg/L in the sailfin molly (Laska *et al.* 1978). The results for 1,1′,2-trichloroethylene produced a 96-hr LC50 of 45.0 mg/L, which compared closely with the 96-hr LC50 of 40.7 mg/L determined for fatheads by Alexander *et al.* (1978). This study yielded a 1,2-dichloropropane fathead 96-hr LC50 of 140 mg/L, whereas Dawson *et al.* (1975, 1977) found a 96-hr LC50 of 320 mg/L for bluegills. Our results produced a 1,2-dichloroethane fathead 96-hr LC50 of 116 mg/L while Dawson *et al.* (1975, 1977) reported a 96-hr LC50 for 550 mg/L for bluegills. When these results are compared to their respective Water Quality Criteria values, it can be seen for example by comparing the tetrachloroethylene 96-hr LC50 (13.4 mg/L) to its Water Quality Criteria chronic effect level (0.84 mg/L) the ratio is 15.9. For 1,1′,2,2′-tetrachloroethane, this proportion is 8.5, pentachloroethane is 6.67, 1,1′,2-trichloroethane is 8.68, hexachlorobutadiene is 10.7, 1,1′,2-trichloroethylene is 2.05 (here there

was no chronic effect level available; adverse behavioral effect level was used instead), 1,2-dichloropropane is 24.6, 1,2-dichloroethane is 5.8, hexachloroethane is 2.80, 1,3-dichloropropane is 23.0. Additional aquatic toxicity data available for these compounds is summarized in Table 1.

References

Adema, D. M. M.: *Daphnia magna* as a test animal in acute and chronic toxicity tests. Hydrobiologia **59,** 125 (1978).

Alexander, H. C., W. M. McCarty, and E. A. Bartlett: Toxicity of perchloroethylene, trichloroethylene, and methylene chloride to fathead minnows. Bull. Environ. Contam. Toxicol. **20,** 344 (1978).

APHA (American Public Health Association), American Water Works Association, and Water Pollution Control Federation: Standard methods for the examination of water and wastewater. 15 ed. Washington, DC: American Public Health Assoc. (1980).

Barrows, M. E., S. R. Petrocelli, K. J. Macek, and J. J. Carroll: Bioconcentration and elimination of selected water pollutants by bluegill sunfish (*Lepomis macrochirus*). Dynamics, exposure and hazard assessment of toxic chemicals, (Pap. Symp.) Meeting date 1978, Pub. by Ann Arbor Sci., Ann Arbor, MI (1980).

Benoit, D. A., F. A. Puglisi, and D. L. Olson: A fathead minnow (*Pimephales promelas*) early life stage toxicity test method evaluation and exposure to four organic chemicals. J. Environ. Pollut. **28,** 189 (1982).

Canton, J. H., and D. M. M. Adema: Reproducibility of short-term and reproduction toxicity experiments with *Daphnia magna* and comparison of the sensitivity of *Daphnia magna* with *Daphnia pulex* and *Daphnia cucullata* in short-term experiments. Hydrobiologia **59,** 135 (1978).

Dawson, G. W., A. L. Jennings, D. Drozdowski, and E. Rider: The acute toxicity of 47 industrial chemicals to fresh and saltwater fishes. J. Hazard. Mater. **1,** 303 (1975/77).

DeFoe, D.: Unpublished data. U.S. Environmental Research Laboratory, Duluth, MN 55804.

Hamilton, M. A., R. C. Russo, and R. V. Thurston: Trimmed Spearman-Karber method for estimating median lethal concentrations in toxicity bioassays. Environ. Sci. Technol. **11,** 714 (1977). Correction **12,** 417 (1978).

Hawley, G. G. (ed.): Condensed chemical dictionary, 9 ed. New York-London: Van Nostrand Reinhold 1977.

Johnson, W. W., and M. T. Finley: Handbook of acute toxicity of chemicals to fish and aquatic invertebrates. U.S. Dept of Interior, Fish and Wildlife Serv., Resource Pub. 137, Washington, DC (1980).

Laska, A. L., C. K. Bartell, D. B. Condie, J. W. Brown, R. L. Evans, and J. L. Laseter: Acute and chronic effects of hexachlorobenzene and hexachlorobutadiene on red swamp crayfish (*Procambrus clarki*) and selected fish species. Toxicol. Applied Pharmacol. **43,** 1 (1978).

LeBlanc, G. A.: Acute toxicity of priority pollutants to water flea (*Daphnia magna*). Bull. Environ. Contam. Toxicol. **24,** 684 (1980).

Mount, D. I., and W. A. Brungs: A simplified dosing apparatus for fish toxicology studies. Water Res. **1,** 21 (1967).

Phipps, G. L., G. W. Holcombe, and J. T. Fiandt: Saturator system for generating toxic water solutions for aquatic bioassays. Prog. Fish-Cult. **44,** 115 (1982).

Richter, J.: Unpublished data. U.S. Environmental Research Laboratory, Duluth, MN 55804.

Rosenberg, R., O. Grahn, and L. Johansson: Toxic effects of aliphatic chlorinated by-products from vinyl chloride production on marine animals. Water Res. **9,** 607 (1975).

Sloof, W.: Detection limits of a biological monitoring system based on fish respiration. Bull. Environ. Contam. Toxicol. **23,** 517 (1979).

U.S. Environmental Protection Agency: Committee on Methods for Toxicity Tests with Aquatic Organisms. Methods for acute toxicity tests with fish, macroinvertebrates, and amphibians. Ecol. Res. Ser., EPA 660/3-75-009 (1975), NTIS No. PB242105 (National Technical Information Service, Springfield, VA 22161).

———: Ambient water quality criteria for chlorinated ethanes. EPA 440/5-80-029 (1980a), NTIS No. PB81-117400.

———: Ambient water quality criteria for dichloropropane and dichloropropene. EPA 440/5-80-043 (1980b), NTIS No. PB81-117541.

———: Ambient water quality criteria for hexachlorobutadiene. EPA 440/5-80-053 (1980c), NTIS No. PB81-117640.

———: Ambient water quality criteria for tetrachloroethylene. EPA 440/5-80-073 (1980d), NTIS No. PB81-117830.

———: Ambient water quality criteria for trichloroethylene. EPA 440/5-80-077 (1980e), NTIS No. PB81-117871.

———: Methods for chemical analysis of water and wastes. Washington, DC: U.S. Environmental Protection Agency, Office of Technology Transfer (1974).

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